in situ hybridization methods. For the Examiner's convenience, the pending claims are provided in Appendix I, below.

Claims 26-41 stand rejected under 35 U.S.C. §112, second paragraph, for allegedly being indefinite. Claims 26, and 45-47 stand rejected under 35 U.S.C. §102(b) for allegedly being anticipated by Zakut-Houri *et al. Nature* 306:594 (1983), Chader *et al.* WO 95/334480, Okubo *et al.* WO 95/14772[1] Morris *et al. Blood* 78:1078-1084 (1991), or Beach WO 93/24514. Claim 45 stands rejected for allegedly being anticipated by either Tanner *et al. Cancer Research* 54:4257 (1994) or Kallioniemi *et al. Proc. Natl. Acad. Sci. USA* 91:2156-2160 (1994).

Response to Restriction

Applicants hereby affirm the oral election of claims 26-41 and 45-47 for prosecution in this application. Contrary, to the statement in the Office Action, however, the election has been made with traverse. According to MPEP §803, if the search and examination of an entire application can be made without serious burden, the Examiner must examine it on the merits, even though it includes claims to distinct or independent inventions. In the present case, applicants respectfully submit that search of all the inventions in the identified groups presents no undue burden. The election is therefore made with traverse.

Rejection under 35 U.S.C. §112, second paragraph

The rejection of claims 26-41 for allegedly being indefinite is overcome by the above-amended claims. In the Office Action, the Examiner has rejected the pending claims because the probes of the invention are allegedly not clearly defined. To expedite prosecution,

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[1] A note apparently written by the Examiner indicates that this disclosure is the same as Okubo *et al. DNA Research* 1:37 (1994). Since the *DNA Research* paper is in English and the published application is in Japanese, applicants rely on the paper for the purposes of this response.

the claims have been amended to incorporate language of the specification defining stringent hybridization conditions for Southern and Northern blots (*see*, page 7, lines 3-14). In particular, claim 26 is now directed to use of probes that hybridize to the exemplified sequences in under stringent conditions that include washing with 0.2x SSC at 60°C for 15 minutes. Support for this amendment is found at page 7, lines 6-7. Withdrawal of the amendment is respectfully requested.

Rejections under 35 U.S.C. §102(b)

The rejection of the claims for allegedly being anticipated by the prior art is respectfully traversed. The Examiner is respectfully reminded that anticipation requires that "all limitations of the claim are found in the reference, or 'fully met' by it." *Kalman v Kimberly-Clark Corp.*, 218 USPQ 781, 789 (Fed. Cir. 1983). As explained below, the cited references fail to anticipate the presently claimed invention because they do not disclose use of nucleic acid probes to detect increased copy number at 20q13.2.

To expedite prosecution, the claims have been amended to recite more specifically that the claimed methods are used to detect increased copy of target sequences in the 20q13.2 amplicon disclosed here. As explained in detail in the specification, amplification in a 2Mb region at 20q13.2 has been correlated with cancer (*see*, page 12, line 22 to page 13, line 30). The present invention is based in part on the discovery and cloning of sequences from particular genes in that region. Sequences from these genes (*e.g.*, cDNA sequences) can be conveniently used to detect the presence of amplification in this region using standard nucleic acid hybridization techniques. As explained below, none of the cited references disclose or suggest that probes specific to the sequences disclosed here could be used to detect amplification in the 20q13.2 amplicon.

Zakut-Houri *et al.* discloses the sequence of the ***mouse*** p53 gene. The Examiner has cited this reference because six nucleotides in the poly A tail of the cDNA sequence in SEQ ID NO:1 are shared by the mouse gene. Applicants respectfully submit that

nothing in this reference discloses or suggests that sequences from this murine gene could be useful in anyway to detect chromosomal abnormalities in human chromosomes. Moreover, it provides no teaching with respect to amplification at 20q13.2. In the absence of such a teaching, the reference cannot anticipate the claimed invention.

Chader *et al.* is cited for teaching the sequence of a gene encoding a pigment epithelium-derived factor (PEDF). The Examiner cites page 11 of this reference for teaching that the gene is detectable in "most tissues, cell types and tumors". Nothing in this reference indicates that this gene is overexpressed or amplified in neoplastic cells. In the absence of evidence that the gene is amplified in cancer, applicants question whether a gene found in most tissues, whether neoplastic or not, is useful to detect neoplastic cells. In addition, nothing in this reference indicates that the gene is located at 20q13.2. In the absence of evidence that the gene is located in this region, there is no motivation for one of skill to use sequences from the gene in the claimed methods.

Okubo *et al.* discloses gene expression profiles in ***normal*** human colonic mucosa. Nothing in this reference indicates that the genes expressed in these cells are correlated in any way to neoplastic growth and would therefor be useful for detection of neoplastic cells. In addition, nothing in this reference indicates that any of the genes are located at 20q13.2. In the absence of evidence that any genes are located in this region, there is no motivation for one of skill to use sequences from the genes in the claimed methods.

Morris *et al.* discloses genes from the chromosomal translocation (9;22) (q34;q11) associated with chronic myelogenous leukemia. Since the translocation involves chromosomes 9 and 22, there is no motivation for one of skill to use sequences from these genes to detect increased copy number at 20q13.2.

Beach *et al.* is cited for teaching a family of cyclin genes that could be associated with cell division associated with neoplastic growth. The Examiner has indicated that one of these genes has sequence similarity to a small part of SEQ ID NO:9. Which gene in Beach *et al.* has this sequence identity is not identified. Three genes are disclosed in the

Beach *et al.* application (*see*, Beach *et al.*, page 5, lines 14-26). Two of these genes have been mapped to 12p13 and 6p21, respectively. No genome location information is provided for the other member of the family. Clearly, one of skill would ***not*** use genes mapped to entirely different chromosomal locations to detect amplification at 20q13.2. With respect to the unmapped gene, there is no reasoning or evidence to show why one of skill would use that gene in the claimed methods.

In light of the above, it is clear that none of the cited references disclose or suggest that the prior art sequences are useful in anyway to detect increased copy number at 20q13.2. Since the claims are clearly directed to detection of sequences from this region, applicants respectfully submit that the rejection of claims 26 and 45-47 should be withdrawn.

On page 6 of the Office Action, claim 45 appears to be separately rejected as being anticipated by either Tanner *et al.* or Kallioniemi *et al.* Claim 45 is directed to methods of detecting overexpression of ***proteins*** encoded by genes in the 20q13 amplicon. One of skill would recognize that overexpression of particular proteins is distinct from copy number increase of genomic sequences at the 20q13 amplicon. For instance, one of skill would realize that detection of overexpression could be carried out by detecting mRNA levels or the protein itself (*see*, specification at page 32, line 10 *et seq.* for a discussion of detecting proteins from the 20q13 amplicon).

As noted above, a rejection based on anticipation is proper only when all limitations of a claim are found in a single prior art reference. Both Tanner *et al.* and Kallioniemi *et al.* are entirely focused on the detection of copy number changes of ***genomic sequences*** at 20q13. Thus, the references cannot anticipate a claim directed to detection of overexpression of genes in that region. The Examiner has identified nothing in these references that indicates that any genes in this region are, in fact, overexpressed. Thus, nothing in these references would lead one of skill to practice the claimed methods.

In view of the foregoing, Applicants believe all claims now pending in this application are in condition for allowance. The issuance of a formal Notice of Allowance at an early date is respectfully requested. If a telephone conference would expedite prosecution of this application, the Examiner is invited to telephone the undersigned at (415) 576-0200.

Respectfully submitted,

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APPENDIX I

26. (Twice amended) A method of screening for neoplastic cells in a sample, the method comprising:

contacting a nucleic acid sample from a human patient with a probe which hybridizes to the target polynucleotide sequence under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes, the target polynucleotide sequence comprising a sequence selected from the group consisting of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, and SEQ ID NO:45 wherein the probe is contacted with the sample under conditions in which the probe hybridizes selectively with the target polynucleotide sequence to form a stable hybridization complex; and

detecting the formation of a hybridization complex, whereby the presence or absence of neoplastic cells having increased copy number at 20q13.2 is determined.

27. The method of claim 26, wherein the nucleic acid sample is from a patient with breast cancer.

28. The method of claim 26, wherein the nucleic acid sample is a metaphase spread or a interphase nucleus.

29. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:1.

30. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:2.

31. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:3.

32. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:4.

33. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:5.

34. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:6.

35. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:7.

36. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:8.

37. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:9.

38. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:10.

39. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:12.

40. The method of claim 26, wherein the probe comprises a polynucleotide sequence as set forth in SEQ ID NO:45.

41. The method of claim 26, wherein the probe is used to identify the presence of a mutation in the target polynucleotide sequence.

45. A method of detecting a cancer, said method comprising detecting the overexpression of a protein encoded in a 20q13 amplicon.

46. The method of claim 45, wherein said protein encoded in a 20q13 amplicon is ZABC1.

47. The method of claim 45, wherein said protein encoded in a 20q13 amplicon is 1b1.

48. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:1 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

49. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:2 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

50. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:3 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

51. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:4 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

52. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:5 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

53. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:6 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

54. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:7 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

55. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:8 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

56. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:9 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

57. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:10 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

58. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:11 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

59. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:12 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

60. The method of claim 26, wherein the probe comprises a polynucleotide sequence that hybridizes to SEQ ID NO:45 under stringent conditions that include washing with 0.2x SSC at 65°C for 15 minutes.

61. The method of claim 26, wherein the probe is labeled.

62. The method of claim 61, wherein the label is a fluorescent label.

63. The method of claim 26, wherein the nucleic acid sample is a chromosome sample.